



Grand challenges in plant biotechnology

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- Increase crop productivity especially in adverse environments
- Management of herbicide tolerance
- Management of resistance to pests
- Management of resistance to diseases
- Improvement of genetic engineering technologies to enhance public perception
- Improvement of harvest index
- Improvement of nutrient cycling in agricultural ecosystems

Since the emergence of agriculture 10,000 years ago after domestication of suitable animals and plants, civilized cultures developed as it was then possible to provide food and feed in surplus allowing additional activities in contrast to societies relying on gathering and hunting. Agricultural practices since then have been continuously developing, not only cultivation practices of plants improved productivity, but also the genetic material used was gradually improved through breeding to sustain ever growing populations. This linear increase in crop productivity however is not satisfying the increased demand anymore, as not only the farmland available per capita is decreasing, but also the demands for renewable products derived from agriculture is increasing as traditionally petroleum-based industries are more and more relying on biomass-derived products, energy being the most prominent example.

The Grand Challenge in Plant Biotechnology therefore lies foremost in increasing crop productivity at orders of magnitude, which has never been achieved so far, but not much less in improving plant quality to be optimal for its traditional uses, e.g., food and feed, but also to provide tailor-made biomaterials for a vast range of industrial applications including the provision of energy for a range of purposes, which can only be achieved if the enabling technologies are also further developed allowing advancements in plant breeding at unprecedented speed.

To this end, it requires a highly interdisciplinary effort to interpret the multi-parallel data sets generated currently and even more so in the future at all levels of plant biology to generate information that can be exploited by plant breeders. Plant biology as compared to human biology is though dealing with the additional level of complexity that is encountered by the fact that plants usually are grown in constantly changing environments that are almost always far from optimal, even more in times where climate is changing gradually in many ecosystems. From the perspective of a plant biologist, this not only requires the integration of specialty fields in plant science, but also the expertise in the broadest range of crop science.

The first contribution of Plant Biotechnology to improve crop productivity was the development of crops that were resistant to broadband herbicides, which are often selective for plants, 15 years ago. These technologies have been proven highly valuable, both at the ecological as well as on the socio-economic level, as less overall and less environmentally questionable herbicides and more economically favorable herbicides are utilized in production systems of such crops. The challenge remains to be able to sustain such production systems with crops resistant to broadband herbicides, as it is foreseeable that after continuous application of such broadband herbicides over a range of growing seasons, that resistance to those herbicides amongst weeds will emerge and the selective advantage will be lost to the crops. The scientific challenge to address this problem will be to diversify the range of broadband herbicides that can be utilized, allowing a crop or variety rotation management that prevents selection for herbicide tolerance in weeds. This not only needs development of novel herbicides targeted to new plant-unique biochemical pathways, but also the discovery of new resistance mechanisms.

A similar challenge resides within in the second trait that has been commercially exploited on a large scale so far, which is resistance to insect pests, and which has been addressed so far by expressing bacterial toxins against the respective pests in crops. Also here a diversification is needed to prevent resistances of the pests to occur. So far only proteinaceous toxins derived from *Bacillus thuringiensis* have been expressed in crops to confer insect pest tolerance however the spectrum of peptides, proteins, or other compounds derived from secondary metabolism needs to be broadened for engineering novel mechanisms of pest resistance allowing an integrated pest management in the future (Kos et al., 2009).

The areas discussed so far have been the almost exclusive ones that have been commercially utilized on the crop level so far, due to the fact that legal and regulatory constraints have been so massive, making the commercialization of genetically engineered crops economically mostly unfeasible for any other field of application, with the exception of specialty applications of plants or plant cell cultures. It remains therefore a challenge for the Plant Science community to be able to influence societies to base their legal frameworks on the commercialization of GM crops and plants on science and not on belief. After 15 years of applying a strictly precautionary principle, it might now be time to re-evaluate matters based on the experiences of risks made. To this end it will be mandatory that Plant Scientists find a forum of communication with colleagues in the broadest fields of Humanities and Laws to be able to influence legislation that is preventing so far that a ground-breaking technology can be utilized to address a range of burning problems for humankind. Public acceptance of this technology however only will grow the more crops are commercialized where the benefit to the consumer or the subsistence farmer are more clearly evident, which will only happen if regulatory demands are loosened

allowing the commercialization of economically less attractive traits. Alternatively, it seems not unlikely that technological improvements allow for applying biotechnological approaches to target genes directly without utilizing antibiotic resistance genes as selectable markers, which might greatly enhance public acceptance. Examples are the selection for knock-out mutants for specific genes using the “targeted induced local lesions in genomes” (TILLING) technology (Colbert et al., 2001), or the application of “intragenesis,” where plants are transformed only with DNA of their origin (Rommens et al., 2007), and, more recently, the utilization of transcription activator like effectors (TALE) has been developed for many different organisms including higher plants to either up-regulate targeted genes (Morbitzer et al., 2010), or to disrupt genes if such effectors are fused with a nuclease (TALEN; Cermak et al., 2011).

Even though knowledge on mechanisms by which plants develop resistance to fungal pathogens has been growing vastly over the last two decades (Schulze-Lefert and Panstruga, 2011) this has only been exploited in plant breeding programs applying marker assisted breeding selecting for quantitative trait loci conferring horizontal disease resistance, as is in general true for quantitative traits. Genetically engineering vertical disease resistance does not have any advantage over traditional breeding unless one is dealing with vegetatively propagated crops, which are usually genetically very complex and traditional inbred selection is impossible to carry out and genetic engineering would therefore be the method of choice to introduce single traits as exemplified by the transgenic papaya that had been engineered to be resistant to papaya ringspot virus and had been introduced to the Hawaiian market in the late 1990s to revitalize the local papaya industry (Gonsalves, 2004). In whatever genetically complex crop it remains the challenge of Plant Biotechnology to discover and engineer mechanisms of disease resistance if bacterial, fungal, or viral diseases emerge that put certain crops representing major industries in areas or even countries under threat.

The major contribution of plant breeding to improving crop yield over the last century mainly lies in, next to adapting crops to novel cultivation practices or develop-

ing mechanisms of disease and pest resistance, improving the harvest index of crops (Gifford and Evans, 1981; Gifford et al., 1984), which is the ratio between biomass produced and biomass harvested. Many attempts have been undertaken for more than 20 years now to engineer carbohydrate partitioning in plants to improve the allocation of assimilates into the harvested organs, but only few examples exist, where this was successfully achieved, namely in potatoes, where it was possible to increase crop yield by elevating the starch content. This was done through manipulation of nucleotide metabolism rather than influencing carbohydrate metabolism directly (Regierer et al., 2002; Geigenberger et al., 2005), and the mechanisms by which the yield increases can be explained still remain elusive (Geigenberger et al., 2009). Understanding the regulation of carbohydrate partitioning between plant organs and generating technology to further improve the harvest index remains a grand challenge. Alternatively it is also conceivable to develop multipurpose crops, where many parts of the crop can be utilized, or where crops are simply engineered, in which the “waste” components have been modified making them more amenable to fermentation (see below). Given the close phylogenetic relation between potato, tobacco, and tomato plants, it is not completely unrealistic to develop plants, which produce edible fruits and tubers, as well as consumable leaves as an example.

In order to satisfy the ever increasing demands to crop production in the context of decreasing resources it becomes of central importance to develop crops that remain being productive with less energy inputs, such as fertilizers, pesticides, and water, or to allow the more efficient recycling of mineral nutrients that are lost with sewage sludge if plants were generated that are not incorporating or not negatively affected by toxic heavy metals or organic compounds contained within the sludge. The scientific challenges pertaining to these issues are more deeply described in the Grand Challenges in Plant Nutrition article by von Wiron (2011), however it should be mentioned here that the biggest dreams of plant biotechnology to serve humankind always has been to enable other plants than legumes to undergo symbiotic relationships allowing for atmospheric nitrogen fixation,

similar to transferring the capability of C4 metabolism to C3 plants. Whether these dreams are realistic remains to be seen, however they also imposed a threat for the public acceptance of plant biotechnology, as these dreams were expressed in the very early stages of this technology, and have not been met by far.

Increasing demands to crop production are continuously imposed by utilizing fatty acids, starch, and sucrose for biofuel production. In order to prevent serious competition to resources needed to satisfy demands for food and feed, it is mandatory to develop alternative bio-based resources for energy production, which do not compete for food and feed. These could be plants that grow in environments, which are currently not cultivated, plants, which produce more energy-dense biomaterials than carbohydrates or fatty acids such as latex (polyterpenoids), which cannot be utilized for nutritional purposes. Latex (natural rubber) is exclusively harvested from the rubber tree, which exclusively grows in tropical climates. Attempts have been undertaken to develop alternative sources of latex, which also grow in more temperate climates, such as guayule (Cornish et al., 1993). This approach seems to be worthwhile to be re-evaluated given the vast biodiversity that remains un-exploited so far. This would also require deeper insights on the processes underlying plant domestication to be able engineer plants, which are promising candidates, into crops.

Finally, there is the ultimate challenge to genetically tailor plant products to optimally suit them as sources of food, feed, fuel, materials, and pharmaceuticals. The ultimate challenge is to understand and influence metabolism without compromising crop productivity, unless high-value compounds such as pharmaceuticals are produced in other plant systems than crops, e.g., in plant cell or tissue cultures.

To meet the challenges a broad interdisciplinary approach needs to be taken next to the scientific and technological prerequisites that have to be met. Multidisciplinarity is not only needed to transfer knowledge generated with model plants into crops, or even highly environmentally specialized varieties, but also to stimulate public acceptance and thus decreasing regulatory restraints. Technologically we will need to be able to generally simplify genetic manipulation of

any plant species, and be able to precisely engineer genomes beyond simply inserting transgenes introducing also a range of traits simultaneously, next to taking maximal advantage of the knowledge generated in any sub-discipline of Plant Science.

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Received: 11 August 2011; accepted: 13 March 2012; published online: 02 April 2012.

Citation: Kossmann J (2012) Grand challenges in plant biotechnology. *Front. Plant Sci.* 3:61. doi: 10.3389/fpls.2012.00061

This article was submitted to *Frontiers in Plant Biotechnology*, a specialty of *Frontiers in Plant Science*.

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